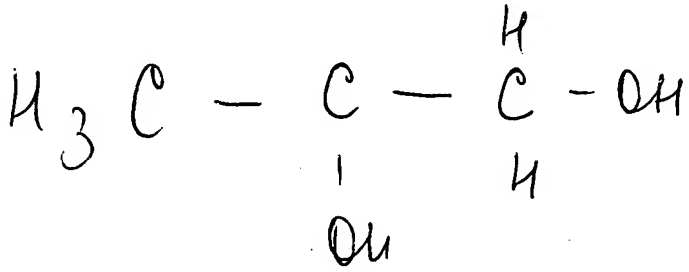
(FILE 'HOME' ENTERED AT 15:51:18 ON 20 JUN 2003)

FILE 'REGISTRY' ENTERED AT 15:51:30 ON 20 JUN 2003
L1 0 S C3 H8 O2
L2 92 S (C3 H8 O2)/MF

L10 ANSWER 92 OF 92 REGISTRY COPYRIGHT 2003 ACS
RN 57-55-6 REGISTRY
CN 1,2-Propanediol (8CI, 9CI) (CA INDEX NAME)
OTHER NAMES:
CN (±)-1,2-Propanediol
CN (±)-Propylene glycol
CN (RS)-1,2-Propanediol
CN a-Propylene glycol
CN 1,2-(RS)-Propanediol
CN 1,2-Dihydroxypropane
CN 1,2-Propylene glycol
CN 1000PG
CN 2,3-Propanediol
CN 2-Hydroxypropanol
CN DL-1,2-Propanediol
CN dl-Propylene glycol
CN Dowfrost
CN Isopropylene glycol
CN Methylethyl glycol
CN Methylethylene glycol
CN Monopropylene glycol
CN PG 12
CN Propylene glycol
CN Sirlene
CN Solar Winter Ban
CN Solargard P
CN Ucar 35
FS 3D CONCORD
DR 63625-56-9, 4254-16-4, 190913-75-8
MF C3 H8 O2
CI COM
LC STN Files: ADISNEWS, AGRICOLA, ANABSTR, AQUIRE, BEILSTEIN*, BIOBUSINESS, BIOSIS, BIOTECHNO, CA, CABA, CANCERLIT, CAOLD, CAPLUS, CASREACT, CBNB, CEN, CHEMCATS, CHEMINFORMRX, CHEMLIST, CHEMSAFE, CIN, CSCHEM, CSNB, DDFU, DETHERM*, DIOGENES, DIPPR*, DRUGU, EMBASE, ENCOMPLIT, ENCOMPLIT2, ENCOMPPAT, ENCOMPPAT2, GMELIN*, HODOC*, HSDB*, IFICDB, IFIPAT, IFIUDB, IPA, MEDLINE, MRCK*, MSDS-OHS, NAPRALERT, NIOSHTIC, PDLCOM*, PHAR, PIRA, PROMT, RTECS*, SPECINFO, SYNTHLINE, TOXCENTER, TULSA, ULIDAT, USAN, USPAT2, USPATFULL, VETU, VTB
(*File contains numerically searchable property data)
Other Sources: DSL**, EINECS**, TSCA**
(**Enter CHEMLIST File for up-to-date regulatory information)



L10 ANSWER 91 OF 92 REGISTRY COPYRIGHT 2003 ACS
RN 109-86-4 REGISTRY
CN Ethanol, 2-methoxy- (8CI, 9CI) (CA INDEX NAME)
OTHER NAMES:
CN b-Methoxyethanol
CN 1-Hydroxy-2-methoxyethane
CN 2-Methoxy-1-ethanol
CN 2-Methoxyethanol
CN 2-Methoxyethyl alcohol
CN 2-Methyloxyethanol
CN 3-Oxa-1-butanol
CN Amsco-Solv EE
CN Dowanol EM
CN Ektasolve EM
CN Ethylene glycol methyl ether
CN Ethylene glycol monomethyl ether
CN Glycol methyl ether
CN Glycol monomethyl ether
CN Methoxyethanol
CN Methoxyethylene glycol
CN Methoxyhydroxyethane
CN Methyl Cellosolve
CN Methyl glycol
CN Methyl oxitol
CN Monoethylene glycol methyl ether
CN Monomethylglycol
CN Poly-Solv EM
FS 3D CONCORD
MF C3 H8 O2
CI COM
LC STN Files: AGRICOLA, ANABSTR, AQUIRE, BEILSTEIN*, BIOBUSINESS, BIOSIS, BIOTECHNO, CA, CANCERLIT, CAOLD, CAPLUS, CASREACT, CBNB, CEN, CHEMCATS, CHEMINFORMRX, CHEMLIST, CHEMSAFE, CIN, CSCHEM, CSNB, DDFU, DETHERM*, DIPPR*, DRUGU, EMBASE, ENCOMPLIT, ENCOMPLIT2, ENCOMPPAT, ENCOMPPAT2, GMELIN*, HODOC*, HSDB*, IFICDB, IFIPAT, IFIUDB, IPA, MEDLINE, MRCK*, MSDS-OHS, NIOSHTIC, PDLCOM*, PHAR, PIRA, PROMT, RTECS*, SPECINFO, TOXCENTER, TULSA, ULIDAT, USPAT2, USPATFULL, VETU, VTB
(*File contains numerically searchable property data)
Other Sources: DSL**, EINECS**, TSCA**
(**Enter CHEMLIST File for up-to-date regulatory information)

L10 ANSWER 89 OF 92 REGISTRY COPYRIGHT 2003 ACS
RN 504-63-2 REGISTRY
CN 1,3-Propanediol (8CI, 9CI) (CA INDEX NAME)
OTHER NAMES:
CN b-Propylene glycol
CN w-Propanediol
CN 1,3-Dihydroxypropane
CN 1,3-Propylene glycol
CN 1,3-Propylenediol
CN 2-Deoxyglycerol
CN PG
CN Trimethylene glycol
FS 3D CONCORD
MF C3 H8 O2
CI COM
LC STN Files: AGRICOLA, ANABSTR, AQUIRE, BEILSTEIN*, BIOBUSINESS, BIOSIS, BIOTECHNO, CA, CANCERLIT, CAOLD, CAPLUS, CASREACT, CBNB, CEN, CHEMCATS, CHEMINFORMRX, CHEMLIST, CIN, CSCHEM, CSNB, DDFU, DETHERM*, DIPPR*, DRUGU, EMBASE, GMELIN*, HODOC*, IFICDB, IFIPAT, IFIUDB, MEDLINE, MRCK*, MSDS-OHS, NAPRALERT, NIOSHTIC, PIRA, PROMT, RTECS*, SPECINFO, SYNTHLINE, TOXCENTER, TULSA, USPAT2, USPATFULL, VTB
(*File contains numerically searchable property data)
Other Sources: DSL**, EINECS**, TSCA**
(**Enter CHEMLIST File for up-to-date regulatory information)

FILE 'REGISTRY' ENTERED AT 15:51:30 ON 20 JUN 2003
L1 0 S C3 H8 O2
L2 92 S (C3 H8 O2)/MF

FILE 'CAPLUS' ENTERED AT 15:53:09 ON 20 JUN 2003
L3 19 S C3H8O2 OR (C3 H8 O2)
L4 29607 S PROPANEDIOL
L5 37501 S SALMONELLA
L6 98 S L4 AND L5
L7 5406 S GRAM NEGATIVE
L8 7 S L7 AND L4
L9 56723 S (BILE OR CHOLIC OR DEOXYCHOLIC)
L10 86 S L4 AND L9
L11 409429 S SILICA
L12 0 S L11 AND L10
L13 319866 S SIO2
L14 0 S L13 AND L10
L15 15136 S SILICEOUS
L16 0 S L15 AND L10
L17 161992 S SILICATE
L18 2 S L17 AND L10
L19 24281 S CREATINE
L20 0 S CULTURE MEDI
L21 65467 S CULTURE MEDI?
L22 1 S L21 AND L19 AND L5
L23 5406 S GRAM NEGATIVE
L24 0 S L23 AND L21 AND L19
L25 48 S L5 AND L19
L26 816361 S MEDIUM OR MEDIA
L27 4 S L26 AND L25
L28 1099 S L26 AND L19
L29 1 S L23 AND L28
L30 30671 S CREATININE
L31 50 S L30 AND L5
L32 3 S L26 AND L31
L33 103138 S CYSTINE OR CYSTEINE
L34 0 S L3 AND L5
L35 0 S L3 AND L26

FILE 'BIOSIS' ENTERED AT 16:13:30 ON 20 JUN 2003
L36 55521 S SALMONELLA
L37 42911 S CREATININE
L38 72 S L37 AND L36
L39 353672 S MEDIA OR MEDIUM
L40 0 S L39 AND L38
L41 166515 S GRAM NEGATIVE
L42 272 S L41 AND L37
L43 13 S L39 AND L42
L44 55668 S CYSTINE OR CYSTEINE
L45 560 S L44 AND L36
L46 119 S L45 AND L39
L47 28 S L41 AND L46
L48 2021 S CYSTEINE AND CYSTINE
L49 20 S L48 AND L36
L50 6 S L49 AND L39

WEST Search History

DATE: Friday, June 20, 2003

Set Name side by side	Query	Hit Count	Set Name result set
	DB=USPT; PLUR=YES; OP=ADJ		
L17	l14 and l16	3	L17
L16	((435/253.6)!.CCLS.)	217	L16
L15	L14 and l8	0	L15
L14	creatinine	3275	L14
L13	L12 and l8	12	L13
L12	cystine	6165	L12
L11	L10 and l8	8	L11
L10	cysteine	27008	L10
L9	L8 and l6	0	L9
L8	((selective or selection oe selecting) with medi?) same salmonella	81	L8
L7	L6 and l5	23	L7
L6	creatinine	3275	L6
L5	l3 and l1	1149	L5
L4	s l3 and l1L3	0	L4
L3	salmonella	11802	L3
L2	salmonella1	0	L2
L1	(selective or selection) with medi?	8597	L1

END OF SEARCH HISTORY

AB Paper chromatographic examn. of the culture medium at different intervals showed some amino acids to disappear while others, newly formed, appeared in the medium. S. paratyphi B formed amino acids from NH_4 salts and P. aeruginosa formed them from either NH_4 salts or urea, but neither organism utilized the N of thiourea, guanidine. or creatinine.

ACCESSION NUMBER: 1958:66916 CAPLUS
DOCUMENT NUMBER: 52:66916
ORIGINAL REFERENCE NO.: 52:12070g-h
TITLE: Biosynthesis of amino nitrogen by Pseudomonas aeruginosa and by Salmonella paratyphi B
AUTHOR(S): Cesaire, O. G.; Neuzil, E.; Boiron, H.; Lecomte, M.
CORPORATE SOURCE: Univ. Dakar, French W. Africa
SOURCE: Comptes Rendus des Seances de la Societe de Biologie et de Ses Filiales (1957), 151, 1167-71
CODEN: CRSBAW; ISSN: 0037-9026
DOCUMENT TYPE: Journal
LANGUAGE: Unavailable

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WEST

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L24: Entry 1 of 2 File: USPT Dec 1, 1998

DOCUMENT-IDENTIFIER: US 5843699 A
TITLE: Rapid microorganism detection method

Brief Summary Text (5):
For example, culturing a food sample for the presence of Salmonella typically involves the addition of approximately 20-25 grams of sample, such as meat, into approximately 225 ml of primary enrichment media. The primary enrichment media is typically non-selective, such as Buffered Peptone Water (BPW) or Universal Pre-enrichment Broth (UPB), to allow for repair of injured microorganisms. The sample is then thoroughly mixed with the primary enrichment broth and incubated for 22-28 hours at approximately 35.degree. C..+-.2.degree. C. Following this step, the sample is further selectively enriched in a growth promoting medium containing inhibitors that allow for the continued growth of a target organism, such as Salmonella, while simultaneously restricting the proliferation of most other competing microorganisms.

Brief Summary Text (6):
The most commonly used method for selective enrichment of the target organisms requires the transfer of approximately one milliliter of the primary enrichment media into two tubes containing ten milliliters of selective media such as Selenite Cystine Broth and Tetrathionate Broth (AOAC), respectively, and incubating these tubes for 22-28 hours at approximately 35.degree. C. Other secondary enrichment media for Salmonella could include Rappaport-Vassiliadis Medium (RV) and Lauryl Tryptose Broth. This secondary enrichment step is generally followed by a detection step that can include plating the secondary enrichment broth onto solid media or by utilizing other more rapid methods, such as immunological assays or DNA probes (all media available from Difco Laboratories, Detroit, Mich.).

Brief Summary Text (16):
Other methods which are described and shown in Table 1, illustrate a number of commercially available test systems for the identification of Salmonella in a sample. All of the methods described in Table 1 require either the plating of selectively grown microorganisms onto a solid selective media to obtain a pure culture or require an immunological or nucleic acid based assay to be performed on every sample in order to obtain a positive or negative result for the presence or absence of Salmonella in the sample.

Detailed Description Text (12):
In the present invention, the pre-enrichment media can also include the addition of a selective inhibitor or mildly selective inhibitor such as an antibiotic. In the method of the present invention, the antibiotic novobiocin can be added to the modified-TSB media prior to the addition and incubation of the sample in order to provide a selection pressure in favor of the growth of Salmonella. The novobiocin or sodium salt thereof can be added in a concentration of between 1-50 .mu.g/ml. Preferably, the concentration of the novobiocin is approximately 20-40 .mu.g/ml with the preferred concentration being approximately 25 .mu.g/ml of pre-enrichment media. Alternatively, other antibiotics such as vancomycin, penicillin, ampicillin, and amikacin can be utilized in appropriate concentrations. Other target organism selection enhancing additives known to those skilled in the art can be added to the pre-enrichment broth to favor the growth of the selected target microorganism over other competing non-target microorganisms present in the sample.

Detailed Description Text (15):

The selective inhibitor(s) or mixture thereof can include compounds or reagents which are known to inhibit the growth of non-target microorganisms while encouraging the proliferation or growth of the target microorganisms. For example, if the target microorganism is Salmonella, it has been found that a mixture of magnesium chloride, malachite green, and crystal violet can be added to the pre-enrichment media present in the container to yield excellent inhibition of non-target microorganisms. The concentration of the malachite green in the solution ranges from approximately 10 mg/l to 50 mg/l (0.003%-0.03 w/v), the magnesium chloride concentration ranges from approximately 0.5 g/l to approximately 2 g/l (0.1%-1.0% w/v), and the concentration of the crystal violet ranges from approximately 0.001 g/l to 0.010 g/l (0.0005%-0.001% w/v). The preferred concentration of the inhibitor mixture for Salmonella includes 30 mg/l malachite green, 0.005 g/l crystal violet, and approximately 1.0 g/l magnesium chloride. Other inhibitors can include bile salts, sodium deoxycholate, sodium selenite, sodium thiosulfate, sodium tetrathionate, sodium sulphacetamide, mandelic acid, selenitecysteine tetrathionate, sulphamethazine, brilliant green, malachite green, crystal violet, tergitol 4, sulphadiazine, amikacin, and novobiocin.

Detailed Description Text (74):
magnesium chloride, anhydrous (Sigma, Lot #86F-3524)

Detailed Description Paragraph Table (13):
TABLE 2
COMPARISON OF THE CONVENTIONAL SALMONELLA TESTING METHOD TO DIFCO'S RAPID SALMONELLA TEST Difco Rapid Competitor Method- Salmonella (Automated Difco Rapid Time Conventional Reveal Transfer) Salmonella (Manual Transfer)

Day 1
Preenrichment: Primary enrichment: Primary enrichment Primary enrichment inoculate and Preenrichment Preenrichment Preenrichment stomach food inoculate and stomach inoculate and stomach inoculate and stomach food sample in non- food sample in non- food sample in media with sample in media with mild inhibitory inhibitory media mild inhibitor inhibitor media Selective enrichment Selective enrichment Selective enrichment incubate 24 add highly selective add highly selective add highly selective hours inhibitors at 4 hours to inhibitors at 4 hours to inhibitors at 4 hours to preenrichment preenrichment preenrichment Secondary enrichment: Secondary enrichment: transfer aliquot to transfer aliquot to reaction me reaction media at 12 hours at 24 hours via manual transfer via automated device Day 2 Selective Detection: Analyze reactions: Analyze reactions: enrichment: immunoassay eliminate 70-90% eliminate 70-90% negative transfer 1 ml all samples need to be negative samples from samples from detection aliquot of tested detection Detection: preenrichment Move to confirmation Detection: immunoassay to selective of immunoassay Move to confirmation of broth media positives Move to confirmation of positives incubate 24 positives hours Day 3 Detection: Plate all samples on selective agar from selective enrichment broth to obtain pure cultures incubate 24 hours Day 4 Analyze plates for suspect Salmonella colonies Move to confirmation of positives

CLAIMS:

15. The method as set forth in claim 1, wherein the inhibitor includes a mixture of magnesium chloride, malachite green, and crystal violet.

32. The system as set forth in claim 29, wherein said growth inhibition means include a mixture of magnesium chloride, malachite green; and crystal violet.

WEST

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L13: Entry 2 of 12 File: USPT Dec 1, 1998

DOCUMENT-IDENTIFIER: US 5843699 A
TITLE: Rapid microorganism detection method

Brief Summary Text (5):
For example, culturing a food sample for the presence of Salmonella typically involves the addition of approximately 20-25 grams of sample, such as meat, into approximately 225 ml of primary enrichment media. The primary enrichment media is typically non-selective, such as Buffered Peptone Water (BPW) or Universal Pre-enrichment Broth (UPB), to allow for repair of injured microorganisms. The sample is then thoroughly mixed with the primary enrichment broth and incubated for 22-28 hours at approximately 35.degree. C..+-.2.degree. C. Following this step, the sample is further selectively enriched in a growth promoting medium containing inhibitors that allow for the continued growth of a target organism, such as Salmonella, while simultaneously restricting the proliferation of most other competing microorganisms.

Brief Summary Text (6):
The most commonly used method for selective enrichment of the target organisms requires the transfer of approximately one milliliter of the primary enrichment media into two tubes containing ten milliliters of selective media such as Selenite Cystine Broth and Tetrathionate Broth (AOAC), respectively, and incubating these tubes for 22-28 hours at approximately 35.degree. C. Other secondary enrichment media for Salmonella could include Rappaport-Vassiliadis Medium (RV) and Lauryl Tryptose Broth. This secondary enrichment step is generally followed by a detection step that can include plating the secondary enrichment broth onto solid media or by utilizing other more rapid methods, such as immunological assays or DNA probes (all media available from Difco Laboratories, Detroit, Mich.).

Brief Summary Text (16):
Other methods which are described and shown in Table 1, illustrate a number of commercially available test systems for the identification of Salmonella in a sample. All of the methods described in Table 1 require either the plating of selectively grown microorganisms onto a solid selective media to obtain a pure culture or require an immunological or nucleic acid based assay to be performed on every sample in order to obtain a positive or negative result for the presence or absence of Salmonella in the sample.

Detailed Description Text (12):
In the present invention, the pre-enrichment media can also include the addition of a selective inhibitor or mildly selective inhibitor such as an antibiotic. In the method of the present invention, the antibiotic novobiocin can be added to the modified-TSB media prior to the addition and incubation of the sample in order to provide a selection pressure in favor of the growth of Salmonella. The novobiocin or sodium salt thereof can be added in a concentration of between 1-50 .mu.g/ml. Preferably, the concentration of the novobiocin is approximately 20-40 .mu.g/ml with the preferred concentration being approximately 25 .mu.g/ml of pre-enrichment media. Alternatively, other antibiotics such as vancomycin, penicillin, ampicillin, and amikacin can be utilized in appropriate concentrations. Other target organism selection enhancing additives known to those skilled in the art can be added to the pre-enrichment broth to favor the growth of the selected target microorganism over other competing non-target microorganisms present in the sample.

Detailed Description Text (19):

The biochemical tests or reagents specific for identification of the target microorganisms can include antibiotics, dyes, and other biochemical reagents indicative of particular microorganisms such as by targeting sugar fermentation, decarboxylation, cleavage by unique enzymes, and/or use of unique combinations of dyes (including fluorescent dyes). Additionally, other reagents used in the detection and identification of microorganisms known to those skilled in the art may be practiced with the present invention. For example, for the detection of Salmonella, the aliquot of the sample from the container can be transferred into a media receiving vessel, such as a test tube, which can contain reagents necessary for performing a MUCAP (methylumbelliferyl caprylate) test. Additional tubes may be utilized which contain reagents for farther biochemical tests such as H.sub.2 S tests, media specifically designed to indicate the presence of lysine decarboxylase, ornithine decarboxylase, or arginine decarboxylase in a suitable base media such as LICNR (lysine-iron-cystine-neurtal red broth, Difco Manual) base media, fermentation reaction media such as for dulcitol, propylene glycol (PG), glucuronic acid (GA), in a peptone base, and a citrate utilization can also be assayed using Simmon's citrate agar (Difco Manual) or citrate medium (magnesium sulfate, ammonium dihydrogen phosphate, sodium citrate, yeast extract, sodium chloride) placed in the media receiving vessels.

Detailed Description Text (106):
disodium L-cystine (Difco, P-2401-20B)

Detailed Description Paragraph Table (2):
Reaction Media: Ingredient g/l
1.) MUCAP + H.sub.2 S Strip Media-citrate based magnesium sulfate 0.2 ammonium dihydrogen 1 phosphate dipotassium phosphate 1 sodium chloride 5 yeast extract 0.3 sodium citrate 5 sodium thiosulfate 0.1 pH of media 7.83 2.) Modified LICNR Media yeast extract 3 tryptone 5 L-lysine 10 L-cystine 0.1 mannitol 5 dextrose 1 salicin 1 neutral red 0.025 pH of media 6.2 +/- 0.2.degree. C. @ 25.degree. C.

Detailed Description Paragraph Table (12):
TABLE 1

SENSI- TIV- ITY/ PRE-ENRICHMENT SELECTIVE STEP ADDITIONAL STEP TOTAL SPECI- SYSTEM NAME INC. TIME/TEMP INC. TIME/TEMP INC. TIME/TEMP ASSAY FORMAT TIME FICITY AOAC

LACTOSE BROTH SELENITE CYSTINE NONE PLATING MEDIA- 3 100% TRYPTONE SOYA BROTH (SC) OR BRILLIANT GREEN BROTH NUTRIENT TETRATHIONATE AGAR HEKTOEN BROTH 24h/3SC BROTH (TT) AGAR XLD AGAR 24h/33C 24h/35C BISMUTH SULFITE 4 DAYS AGAR 48h/35C ISO BUFFERED PEPTONE RAPPAPORT- NONE PLATING MEDIA- 3 100% WATER (BPW) VASSILIADIS BRILLIANT GREEN 18h/35-37C BROTH (RV) AGAR 24h/35C 8h/42C OR SC BROTH 8h/35C SALMONELLA-TEK LACTOSE BROTH TT BROTH POST-ENRICHMENT: ELISA- 4 DAYS ORGANON- NUTRIENT BROTH 18-24h/42C AND TRANSFER FROM MONOCLONAL TEKNIKA 24+/- 2h/35C SC BROTH SELECTIVE MEDIUM ANTIBODIES 96.5% CORPORATION 18-24h/35C TO M-BROTH WITH APPROX. 2 hrs 89.3% 10 ug/ml NOVOBIOCIN 16-20h/35C TECRA NON-INHIBITORY TT BROTH AND POST-ENRICHMENT: ELISA- 3 DAYS INTERNATIONAL BROTH SC BROTH TRANSFER FROM SALMONELLA 98.6% BIOPRODUCTS, INC 18-22h/35C 6-8h/35C SELECTIVE MEDIUM VISUAL 96.1% TO M-BROTH IMMUNOASSAY 16-20h/35C APPROX. 2 hrs TECRA UNIQUE RAW FLESH FOODS: NONE- NONE ELISA- 2 DAYS INTERNATIONAL MODIFIED BPW WITH SELECTIVE DIPSTICK ASSAY BIOPRODUCTS, INC TERGITOL-7 OTHER THROUGH DIPSTICK APPROX. 6 hrs FOODS: MODIFIED BPW 16h/35C TECRA MODIFIED BPW WITH IMMUNOENRICH- NONE ELISA- 2 DAYS SALMONELLA EXTRA PHOSPHATE MENT:1 TUBE DIPSTICK ASSAY IMMUNOCAPTURE BUFFER MODIFIED BPW 1 APPROX. 6 hrs INTERNATIONAL MIN 16h/35-37C TUBE OF M-BROTH BIOPRODUCTS, INCUBATE WITH INC. DIPSTICKS AT ROOM TEMP FOR A MINUMUM OF 20 MIN PATH STIK BP WATER OR RV BROTH POST-ENRICHMENT: ELISA- 3 DAYS LUMAC BV BP WATER WITH 16-24h/42C BP WATER DIPSTICK ASSAY 93.0% BRILLIANT GREEN 6-8h/37C 10 MINUTES 96.4% 16-24H/37C 1-2 TEST LOW MICRO LOAD: NONE NONE IMMUNODIFFUSION- 3 DAYS BIOCONTROL NONINHIBITORY WHITE BAND OF PPT SYSTEMS, INC BROTH 24h/35C RESULTS FROM HIGH MICRO LOAD: HIGH MICRO LOAD: IMMOBILIZATION 85- PRE-ENRICH AS IN IODINE ACTIVATED OF MOTILE 100% BAM 24h/35C TETRATHIONATE SALMONELLA 100% BRILLIANT THROUGH SEMI- GREEN BROTH SOLID MEDIUM 8h/42C 16-50h/35C BIO MERIEUX VIDAS BIO MERIEUX MODIFIED ISO BP WATER RV MEDIA METHOD 18h/35-37C 6-8h/42C AND SC BROTH POST-ENRICHMENT: ELFA- 3 DAYS 6-8h/35-37C TRANSFER FROM ENZYME-LINKED NON-SELECTIVE SELECTIVE MEDIUM FLUORESCENT MODIFIED BAM MEDIUM TT BROTH TO M-BROTH IMMUNO/ASSAY METHOD 18h/35-37C 6-8h/42C AND 18h/42C 45 MINUTES SC BROTH 6-8h/35-37C GENE TRAK RAW MEATS: SC BROTH POST-ENRICHMENT:

NUCLEIC ACID- RAW GENE TRAK LACTOSE BROTH 16-18h/35C TRANSFER FROM BASED ASSAY MEATS: SYSTEMS 22-24h/35C SELECTIVE MEDIUM APPROX. 2hrs 4 98.5% OTHER FOODS: TT BROTH AND TO GRAM-NEGATIVE OTHER 97.5% BAM/AOAC METHOD SC BROTH BROTH FOODS: 22-24h/35C 6h/35C 12-18h/35C 3 DAYS DYNABEAD BP WATER IMMUNOMAGNETIC NONE PLANTING MEDIA- 3 DAYS DYNAL AS 16-20h/37C SEPARATION (IMS): XLD AND BGA SELECTIVE BEADS 24h/37C ARE COMBINED WITH PREENRICHMENT 10 MIN AMBIENT TEMP MICRO-SCREEN NEOGEN CORP METHOD A REVIVE MEDIUM SC BROTH NONE CHROMATOGRAPHY 2 DAYS 24h/37C 18h/45C COMBINED WITH GOLD LABELED IMMUNOSORBENT ASSAY (GLISA) APPROX. < 8h METHOD B BAM METHOD SC AND TT BROTH ISOLATION ON 4 DAYS LACTOSE BROTH 24h/37C SELECTIVE AGAR 22-26h/37C AND MEDIA RV BROTH 24h/35C 24h/44C

Detailed Description Paragraph Table (13):

TABLE 2

COMPARISON OF THE CONVENTIONAL SALMONELLA TESTING METHOD TO DIFCO'S RAPID SALMONELLA TEST Difco Rapid Competitor Method- Salmonella (Automated Difco Rapid Time Conventional Reveal Transfer) Salmonella (Manual Transfer)

Day 1

Preenrichment: Primary enrichment: Primary enrichment Primary enrichment inoculate and Preenrichment Preenrichment Preenrichment stomach food inoculate and stomach inoculate and stomach inoculate and stomach food sample in non- food sample in non- food sample in media with sample in media with mild inhibitory inhibitory media mild inhibitor inhibitor media Selective enrichment Selective enrichment Selective enrichment incubate 24 add highly selective add highly selective add highly selective hours inhibitors at 4 hours to inhibitors at 4 hours to inhibitors at 4 hours to preenrichment preenrichment preenrichment Secondary enrichment: Secondary enrichment: transfer aliquot to transfer aliquot to reaction me reaction media at 12 hours at 24 hours via manual transfer via automated device Day 2 Selective Detection: Analyze reactions: Analyze reactions: enrichment: immunoassay eliminate 70-90% eliminate 70-90% negative transfer 1 ml all samples need to be negative samples from samples from detection aliquot of tested detection Detection: preenrichment Move to confirmation Detection: immunoassay to selective of immunoassay Move to confirmation of broth media positives Move to confirmation of positives incubate 24 positives hours Day 3 Detection: Plate all samples on selective agar from selective enrichment broth to obtain pure cultures incubate 24 hours Day 4 Analyze plates for suspect Salmonella colonies Move to confirmation of positives

STIC-ILL 10908280 451765 Main/pmc SF224.J86

From: Afremova, Vera
Sent: Friday, June 20, 2003 6:04 PM
To: STIC-ILL
Subject: 10/018,738

NOS
6/23/03
SA

Hi, please, could I have these references::

1
ACCESSION NUMBER: 1993:501200 BIOSIS
TITLE: Iontophoresis and chloride-containing compounds: Parameters required for killing.
AUTHOR(S): Davis, C. P. (1); Wagel, N.; Anderson, M. D.; Warren, M. M.
SOURCE: Journal of Urology, (1993) Vol. 150, No. 4, pp. 1172-1175.

2.
ACCESSION NUMBER: 1999:100256 BIOSIS
TITLE: Relative effectiveness of selenite cystine broth, tetrathionate broth, and Rappaport-Vassiliadis medium for the recovery of Salmonella spp. from foods with a low microbial load.
AUTHOR(S): Hammack, Thomas S. (1); Amaguana, R. Miguel; June, Geraldine A.; Sherrod, Patricia S.; Andrews, Wallace H.
SOURCE: Journal of Food Protection, (Jan., 1999) Vol. 62, No. 1, pp. 16-21.

COMPLETED

Vera Afremova
CM1 11E13
308-9351

9921822

Journal of Food Protection, Vol. 62, No. 1, 1999, Pages 16–21

Relative Effectiveness of Selenite Cystine Broth, Tetrathionate Broth, and Rappaport-Vassiliadis Medium for the Recovery of *Salmonella* spp. from Foods with a Low Microbial Load

THOMAS S. HAMMACK,* R. MIGUEL AMAGUAÑA, GERALDINE A. JUNE, PATRICIA S. SHERROD, AND WALLACE H. ANDREWS

Division of Microbiological Studies, U.S. Food and Drug Administration, 200 C Street, SW, HFS-516, Washington, D.C. 20204, USA

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ABSTRACT

The relative effectiveness of Rappaport-Vassiliadis (RV) medium, selenite cystine (SC) broth, and tetrathionate (TT) broth for the recovery of *Salmonella* spp. from foods with a low microbial load was determined. RV medium made from its individual ingredients and incubated at 42°C was compared with a commercial preparation of SC broth, incubated at 35°C, and TT broth incubated at 35 and 43°C, for the recovery of *Salmonella* spp. Twenty-one artificially contaminated food types that included dairy foods, spices, and egg products, as well as other low-microbial-load foods, were analyzed. The foods were inoculated with single *Salmonella* serovars at target levels ranging from 0.04 to 0.4 CFU/g. No significant differences ($P \leq 0.05$) among the selective enrichment broths for the recovery of *Salmonella* spp. from 18 of the foods were observed. Significantly fewer *Salmonella*-positive test portions of gelatin, guar gum, and nonfat dry milk were recovered with RV medium than with SC broth incubated at 35°C and TT broth incubated at 35 and 43°C. TT broth incubated at 35°C recovered the greatest number of *Salmonella*-positive test portions. For the recovery of *Salmonella* spp. from foods with a low microbial load, it is recommended that TT broth incubated at 35°C and RV medium incubated at 42°C be used.

The *Bacteriological Analytical Manual* (BAM) *(23)* and the AOAC *Official Methods of Analysis* (AOAC) *(7)* culture methods for *Salmonella* spp. specify preenrichment, selective enrichment, and selective plating for the recovery of *Salmonella* spp. from foods. The selective enrichment media specified for use with foods with a low microbial load are selenite cystine (SC) and tetrathionate (TT) broths incubated for 24 h at 35°C *(7, 23)*. In previous editions of the BAM, these media were specified for use with all foods. Rappaport-Vassiliadis (RV) medium has consistently been reported to be superior to SC and TT broths for the recovery of *Salmonella* spp. from raw flesh and other highly contaminated foods *(2, 3, 8, 9, 15–17, 19, 22, 24–28)*. In addition, TT broth incubated at 43°C was reported to be superior to TT broth incubated at 35°C *(6, 11, 13, 15, 16, 21)*. Precollaborative and collaborative studies conducted by this laboratory demonstrated the superiority of RV medium incubated at 42°C and of TT broth incubated at 43°C over SC and TT broths incubated at 35°C for the recovery of *Salmonella* spp. from raw flesh and other highly contaminated foods *(15, 16)*. On the basis of these results, the selective enrichment media recommended by the BAM and the AOAC culture methods for *Salmonella* spp. in raw flesh and other highly contaminated foods were changed from SC and TT broths incubated at 35°C to RV medium and TT broth incubated at 42 and 43°C, respectively *(7, 23)*.

The superiority of RV medium over SC broth for the recovery of *Salmonella* spp. from highly contaminated foods *(2, 3, 15, 16, 19, 24)* indicates that, for the sake of method uniformity, it may be appropriate to replace SC broth with RV medium for foods with a low microbial load. The toxicity of SC broth is a further justification for its replacement. Spent SC broth contains toxic levels of selenium, which increases the cost of its disposal because it is classified as hazardous waste by the Environmental Protection Agency *(10)*. Moreover, selenium has been reported to be embryotoxigenic and teratogenic in animal studies *(14, 29)*. It has also been reported to be neurotoxigenic, nephrotoxigenic, hepatotoxigenic, and splenotoxigenic in humans, as well as causing "blind staggers" and "alkali disease" in livestock *(29)*.

The enhanced recovery of *Salmonella* spp. from highly contaminated foods by TT broth incubated at 43°C *(6, 11, 13, 15, 16, 21)* indicates that it would be worthwhile to investigate the possibility of increasing the temperature of TT broth to 43°C for foods with a low microbial load as well. This change, in combination with the possible replacement of SC broth with RV medium, would lead to a single BAM-AOAC selective enrichment procedure for all foods. Statistical equivalence between SC broth and RV medium would allow for the replacement of SC broth with RV medium. Similarly, statistical equivalence between TT broth incubated at 35°C and TT broth incubated at 43°C would allow for the elevation of the TT broth incubation to 43°C as well.

In this study, the relative efficiencies of SC broth incubated at 35°C, TT broth incubated at 35°C and 43°C, and RV medium incubated at 42°C were compared.

* Author for correspondence. Tel: 202-205-4753; Fax 202-401-7740; E-mail: tsh@cfsan.fda.gov.

METHODS AND MATERIALS

Sources of foods. Foods were either purchased in bulk from wholesale outlets located throughout the United States or purchased from retail outlets located in the Washington, D.C., area.

Source of inocula. *Salmonella* spp. cultures were obtained from the stock culture collection of the Division of Microbiological Studies, U.S. Food and Drug Administration. The *Salmonella* spp. cultures had all been isolated from regulatory food samples.

Preparation of inocula. Brain heart infusion (BHI) agar slants were inoculated with *Salmonella* spp. and incubated for 18 to 24 h at 35°C. BHI broth was inoculated from these slants and incubated for 18 to 24 h at 35°C. Ten-milliliter aliquots from the broth were centrifuged at 3,090 × *g*, and the pellets were suspended in 10 ml of Butterfield's phosphate buffer (BPB; pH 6.8 to 7.2). The cells were washed twice with 10-ml aliquots of BPB, and 10-fold serial dilutions were performed. The diluted cell suspensions were lyophilized for inoculation into dried foods or were directly inoculated into nondried foods. Culture suspensions were prepared for lyophilization by mixing 2.5-ml aliquots from appropriate serial dilutions with 2.5-ml aliquots of double strength instant nonfat dry milk (20%, wt/vol), shell freezing the suspensions on an automatic shell freezer in ethanol at −60°C, and lyophilizing the suspensions for 24 ± 2 h in a freeze dryer (VirTis Co., Gardiner, N.Y.).

Inoculation of dried test foods. The lyophilized cultures were ground to a fine powder with a mortar and pestle and mixed with approximately 50 g of uninoculated processed food. The 50-g seed inoculum was mixed with 1.3 to 3.0 kg of the food and aged for a minimum of 2 weeks at room temperature. Bulk foods were inoculated with culture dilutions estimated to give target contamination levels of 0.04 and 0.4 *Salmonella* CFU per g of test food on the day of initiation of analysis.

Inoculation of other test foods. Pecans, egg noodles, peanut butter, and milk chocolate were directly inoculated with cell suspensions estimated to give target contamination levels of 0.04 and 0.4 *Salmonella* CFU per g of test food on the day of initiation of analysis.

Pecan halves (1.5 kg) were immersed in an inoculum bath, stirred for 15 min, drained, and air dried for 30 min at room temperature. The pecans were stored for approximately 10 days at 0 to 5°C before initiation of analysis.

Egg noodles (1.5 kg) were immersed in an inoculum bath, stirred intermittently for 15 min, drained, and frozen overnight to −30°C. The frozen noodles were placed in a freeze dryer, cooled to a temperature of −50°C, and lyophilized for 3 to 4 days depending on their water content. The lyophilized egg noodles were stored a minimum of 2 weeks at room temperature before initiation of analysis.

Peanut butter (2.3 to 3.6 kg) was inoculated, stirred for 15 min, and stored for a minimum of 10 days at room temperature before initiation of analysis.

Milk chocolate (1.5 kg) was heated, with intermittent stirring, in a steam bath until it was fully melted. Sterile distilled water was added, as needed, to give the melted chocolate a smooth consistency. The chocolate was tempered to 60°C and inoculated. The melted inoculated chocolate was stirred with a hand blender for 5 min to achieve homogeneity, spread onto sterile aluminum foil, and allowed to harden at room temperature. It was stored at room temperature for a minimum of 2 weeks before initiation of analysis.

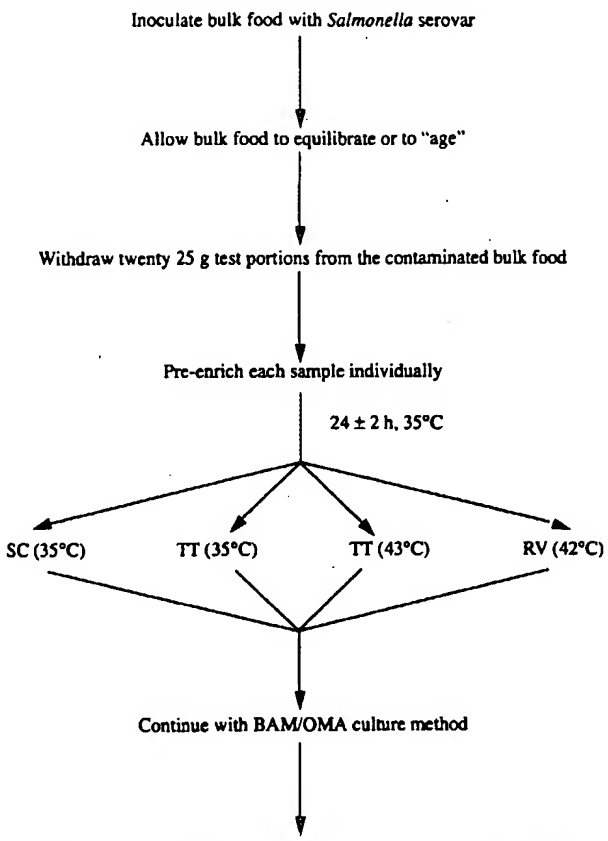

FIGURE 1. *Protocol for comparing relative effectiveness of selenite cystine broth, tetrathionate broth, and Rappaport-Vassiliadis medium for recovery of* Salmonella *from foods with a low microbial load. BAM,* Bacteriological Analytical Manual; *OMA,* Official Methods of Analysis.

Examination of processed foods. Twenty 25-g test portions were withdrawn from the bulk contaminated food and preenriched at a 1:9 test portion-to-broth ratio for 24 ± 2 h at 35 ± 2°C *(7, 23)* (Fig. 1). Gelatin was added to lactose broth containing papain (final concentration 0.1%, wt/vol), at a 1:9 test portion-to-broth ratio and mixed by swirling *(5)*. The gelatin was incubated for 1 h at 35°C, the pH was adjusted to 6.8 ± 0.2, and the gelatin was incubated for an additional 24 ± 2 h at 35°C. Five 25-g uninoculated control test portions were also preenriched. From the incubated preenrichments, 1-ml aliquots were subcultured to 10-ml portions of SC and TT broths and incubated for 24 ± 2 h at 35 ± 2°C; 1-ml aliquots were subcultured to 10-ml portions of TT broth and incubated for 24 ± 2 h at 43 ± 0.2°C; and 0.1-ml aliquots were subcultured to 10-ml portions of RV medium and incubated for 24 ± 2 h at 42 ± 0.2°C. Each incubated selective enrichment was streaked to bismuth sulfite (BS), Hektoen enteric, and xylose lysine desoxycholate agar plates which were incubated for 24 ± 2 h at 35 ± 2°C. Presumptively positive colonies on the selective agar plates were picked to triple sugar iron agar (TSI) and lysine iron agar slants and incubated for 24 ± 2 h at 35 ± 2°C. BS agar plates that contained no presumptive positive colonies after 24 ± 2 h were incubated for an additional 24 h at 35 ± 2°C. Growth from presumptively positive TSI slants was confirmed as *Salmonella* spp. with somatic group antisera (Difco Laboratories, Detroit, Mich.)

TABLE 1. *Relative effectiveness of selenite cystine broth, tetrathionate broth, and Rappaport-Vassiliadis medium for the recovery of* Salmonella *spp. from foods with a low microbial load*

Food	Serovar	MPN[a]	Number of *Salmonella*-positive replicates 20 replicates			
			SC(35)[b]	TT(35)	TT(43)	RV(42)
Noninstant nonfat dry milk	*S.* Infantis	0.93	20	20	20	20
		0.231	20	20	20	20
		0.092	19	19	19	20
		0.04	20	20	20	18
Instant nonfat dry milk						
Brand A	*S.* Miami	0.231	20	20	20	20
		0.092	18	19	20	20
	S. Senftenberg	0.231	13A[c]	13A	14A	8B
		0.043	13	13	13	11
		0.014	8	8	9	5
		<0.003	2	3	3	3
Brand B	*S.* Miami	0.092	20	20	20	20
		0.004	1	1	1	1
	S. Montevideo	0.231	19	20	20	19
	S. Senftenberg	0.021	14	17	18	17
Dry whole milk	*S.* California	0.231	20	20	20	20
		0.093	11	11	10	11
Lactic casein	*S.* Muenchen	0.023	14	14	14	10
		0.023	7	7	7	7
Rennet casein	*S.* Bareilly	0.427	20	20	20	20
		0.009	6	6	6	6
		<0.003	2	2	2	2
Infant formula	*S.* Poona	0.064	19	19	17	18
		0.023	13	13	13	13
Soy flour	*S.* Gaminara	0.93	20	18	20	20
		0.15	19	19	19	19
Cake mix	*S.* Rubislaw	2.4	19	20	20	20
		0.023	7	9	9	9
Inactive dry yeast	*S.* Heidelberg	1.5	20	20	20	20
		0.23	20	20	20	20
		0.23	20	20	20	20
		0.042	17	17	18	17
		0.009	1	1	1	1
		<0.003	1	1	1	0
Active dry yeast	*S.* Illinois	1.49	20[d]	20	20	20
		0.231	20[d]	20	20	20
		0.074	19[d]	19	19	19
		0.042	19[d]	18	18	17
Onion powder	*S.* Miami	0.093	19	19	18	19
		0.043	14	13	14	11
Garlic powder	*S.* Cubana	0.427	20	20	20	20
		0.231	20	20	20	20
Black pepper	*S.* Newbrunswick	0.093	19	20	20	20
		0.015	13	13	13	13
Guar gum						
Brand A	*S.* Montevideo	0.036	11A	11A	9A	4B
		0.03	9A	9A	7AB	4B
	S. Typhimurium	0.231	20A	20A	19AB	15B
		0.072	5	6	4	4
		<0.003	9	9	7	5
		<0.003	3	3	2	0
Brand B	*S.* Montevideo	2.4	20A	20A	19A	13B
		0.147	11A	14A	11A	2B
	S. Typhimurium	0.231	20	20	20	20
		0.036	9B	14A	13AB	13AB
Peanut butter	*S.* Javiana	0.24	20	20	20	20
		0.015	8	8	8	8
		0.004	1	1	1	1

TABLE 1. *Continued.*

Food	Serovar	MPN[a]	Number of *Salmonella*-positive replicates 20 replicates			
			SC(35)[b]	TT(35)	TT(43)	RV(42)
Gelatin						
Brand A	*S.* Anatum	0.074	4	4	5	4
		0.043	13A	13A	13A	8B
Brand B	*S.* Anatum	0.231	20	20	20	20
		0.074	19	19	19	19
		0.009	4	4	4	3
Pecans	*S.* Siegburg	0.427	19	19	19	19
		0.042	4	4	4	4
		0.004	2	2	2	1
Egg yolk (dried)	*S.* Enteritidis	0.427	20	20	20	20
		0.043	15	15	15	15
Egg noodles (dried)	*S.* Alachua	0.093	19	19	19	19
		0.023	12	12	12	12
		0.009	1	1	1	1
Milk chocolate	*S.* Kentucky	4.62	20	20	20	20
		0.427	19	19	19	19
		0.004	4	4	4	4
Coconut	*S.* Madelia	0.231	20	20	20	20
		0.038	3	3	3	3
Total number of positive test portions[c]			1,010	1,025	1,015	954

[a] Most probable number per g of food.
[b] SC, selenite cystine broth; TT, tetrathionate broth; RV, Rappaport-Vassiliadis medium.
[c] Values within a row not sharing a common letter are significantly different ($P \leq 0.05$).
[d] Lauryl tryptose broth.
[e] Total number of *Salmonella*-positive test portions by individual set of medium and incubation temperature conditions.

Determination of levels of inoculation. A three-tube most probable number (MPN) analysis *(23)* was performed to determine the level of contamination in the bulk food on the day of initiation of analysis. MPN portions (100, 10, and 1 g) from the food were rehydrated at a 1:9 ratio of test portion to preenrichment medium by blending, swirling, or soaking, as recommended by the BAM *(23)*.

For food types that were blended, 100-g MPN portions of foods were divided into two 50-g portions that were each blended with 450 ml of preenrichment media and recombined in a single 2-liter Erlenmeyer flask; the 10-g MPN portions of foods were prepared by withdrawing three 100-ml aliquots from a 450-ml blended homogenate and placing each of the three aliquots in individual 250-ml Erlenmeyer flasks; the 1-g MPN portions of foods were prepared by withdrawing three 10-ml aliquots from a 450-ml blended homogenate and placing each of the three aliquots into individual test tubes (16 by 150 mm); the 0.1-g MPN portions of foods were prepared by withdrawing three 1-ml aliquots from a 450-ml blended homogenate and by adding each of the aliquots to three 9-ml portions of preenrichment media contained in individual test tubes (16 by 150 mm).

Foods prepared for examination by swirling or soaking were prepared in a similar fashion except that the 0.1-g MPN portions of foods were added to 9.9 ml of preenrichment media.

MPN preenrichments were incubated for 24 ± 2 h at 35 ± 2°C. One-milliliter portions from each incubated MPN preenrichment were subcultured to 10-ml portions of SC and TT broth and incubated for 24 ± 2 h at 35 ± 2°C. Selective enrichment media were streaked to selective agar plates, and presumptively positive isolates were confirmed as previously described.

Media and reagents. All media and reagents, except for the preenrichment media used with gelatin and guar gum, were prepared according to AOAC Official Methods 967.25 and 967.28 *(7)*. For guar gum, the preenrichment medium specified by the BAM *(4, 23)* was used. For gelatin, the preenrichment medium used was lactose broth with papain (final concentration 0.1% wt/vol) *(5)*.

Statistical analysis. Data from individual experiments were analyzed using SAS software (SAS Institute, Cary, N.C.) with McNemar's chi-square test ($P \leq 0.05$) *(20)*. Combined data, from multiple experiments, were analyzed using SAS software with Duncan's multiple range test ($P \leq 0.05$) *(12)*.

RESULTS AND DISCUSSION

No significant differences among the media for the recovery of *Salmonella* spp. from 18 of the 21 processed foods were observed (Table 1). However, significantly fewer *Salmonella*-positive test portions from brands A and B of guar gum inoculated with *Salmonella* Montevideo, were recovered with RV medium than with SC and TT broths. Moreover, significantly fewer *Salmonella* Typhimurium-positive test portions were recovered with RV medium than were recovered with SC and TT broths from brand A of guar gum. The fact that these differences occurred with two different brands of guar gum and with two different *Salmonella* serovars indicates the differences were neither brand nor serovar dependent. The cause of these differences was not readily apparent.

TABLE 2. *Relative effectiveness of the individual use of selenite cystine broth, tetrathionate broth, and Rappaport-Vassiliadis medium for the recovery of* Salmonella *spp. from foods with a low microbial load*

Food type	Total number of positive test portions[a]	Number of positive test portions for each set of enrichment and temperature conditions			
		SC(35)[b]	TT(35)	TT(43)	RV(42)
All foods	1,055	1,010A[c]	1,025A	1,015A	954B
All foods except guar gum	924	893AB	899A	904A	874B
Guar gum	131	117A	126A	111A	80B

[a] Total number of test portions that were *Salmonella*-positive from any one or more of the four sets of selective enrichment and temperature conditions.
[b] SC, selenite cystine broth; TT, tetrathionate broth; RV, Rappaport-Vassiliadis medium.
[c] Values within a row not having a common letter are significantly different ($P \leq 0.05$).

There were also significant differences among the media for the recovery of *Salmonella* spp. from gelatin and instant nonfat dry milk (Table 1). With both food types significantly fewer *Salmonella*-positive test portions were recovered with RV medium than with the other selective enrichment media. These differences represented singular events among multiple experiments that examined the effects of both food brand and serovar variation for the recovery of *Salmonella* spp. from foods. These differences do not appear to indicate a major deficiency in RV medium that would contraindicate its use with either of these two foods.

There was no overall significant difference between RV medium and SC broth for the recovery of *Salmonella* spp. from 20 of the 21 foods examined, excluding guar gum (Table 2). Moreover, data in Table 1 show that for 18 of the foods examined, the recoveries of *Salmonella* spp. by the selective enrichment media were virtually identical. The significant difference, shown in Table 2, between RV medium and the other selective enrichment media was due to the poor recovery of *Salmonella* spp. from guar gum. These results indicate that SC broth can be replaced with RV medium for the recovery of *Salmonella* spp. from foods with a low microbial load.

Because the BAM recommends the use of selective enrichment pairs, it is also useful to compare the relative effectiveness of the current selective enrichment pair with its possible successors. The relative effectiveness of the selective enrichment combinations TT (35°C)-SC (35°C), TT (35°C)-RV (42°C), and TT (43°C)-RV (42°C) is shown in Table 3. The TT (35°)-TT (43°C) selective enrichment combination was not included because of the possibility that a particular selective enrichment broth may inhibit the growth of some *Salmonella* spp. *(1, 18)*, thus necessitating the use of a second selective enrichment broth to broaden the coverage for *Salmonella* spp. There was no significant difference between the selective enrichment combinations TT (35°C)-SC (35°C) and TT (35°C)-RV (42°C) for the combined data from all of the low microbial load foods examined, including guar gum. All three selective enrichment combinations were equivalent for the recovery of *Salmonella* spp. from 20 of the 21 foods examined, excluding guar gum. Moreover, there was no significant difference between TT (35°C)-SC (35°C) and TT (35°C)-RV (42°C) for the recovery of *Salmonella* spp. from guar gum. Since TT (35°C)-SC (35°C) and TT (35°C)-RV (42°C) were equivalent in all cases and the use of TT (35°C)-RV (42°C) was not contraindicated for use with any food, the selective enrichment combination TT (35°C)-RV (42°C) can be used for the recovery of *Salmonella* spp. from foods with a low microbial load.

The superiority of RV medium and TT broth incubated at 42 and 43°C, respectively, shown by precollaborative and collaborative studies conducted by this laboratory for the recovery of *Salmonella* spp. from foods with a high microbial load *(15, 16)*, was not similarly demonstrated with the foods examined here. This apparent lack of superiority may have been due to the relatively low numbers of competitors

TABLE 3. *Relative effectiveness of selective enrichment media combinations for the recovery of* Salmonella *spp. from artificially contaminated foods with a low microbial load*

Food type	Total number of positive test portions[a]	Number of positive test portions for each combination of enrichment and temperature conditions		
		TT(35)-SC(35)[b]	TT(35)-RV(42)	TT(43)-RV(42)
All foods	1,055	1,038A[c]	1,031A	1,019B
All foods except guar gum	924	905A	905A	905A
Guar gum	131	131A	126A	111B

[a] Total number of test portions that were *Salmonella*-positive from any one or more of the four sets of selective enrichment and temperature conditions.
[b] SC, selenite cystine broth; TT, tetrathionate broth; RV, Rappaport-Vassiliadis medium.
[c] Values within a row not having a common letter are significantly different ($P \leq 0.05$).

present in the foods. Aerobic plate counts for the foods used in this study ranged from 10 to 10^3 CFU/g, with the exception of black pepper, for which the CFU level was 10^5/g. Aerobic plate counts for foods with a high microbial load, used in earlier studies, ranged from 10^4 to 10^9 CFU/g. Where the enhanced selectivity of RV medium and TT broth incubated at 43°C had been shown to increase the recovery of *Salmonella*-positive test portions from highly contaminated bulk foods by inhibiting the growth of competitors in favor of *Salmonella* spp., the relative absence of competitors in processed foods eliminates the apparent advantage of increased selectivity since selectivity requires the presence of competitors to be observed. Thus, the equivalence among the media demonstrated in this study, rather than the superiority of RV medium and TT broth incubated at 43°C shown in earlier studies, was to be expected.

In conclusion, SC broth can be replaced with RV medium, and the selective enrichment media combination of TT (35°)-RV (42°) is the most effective replacement for the current enrichment combination of TT (35°)-SC (35°) for use with foods with a low microbial load.

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